diet (CON: 50% barley silage, and 50% concentrate mix on DM basis), the diet partially replacing barley silage with DDGS (DG: 30% barley silage, 20% DDGS, and 50% concentrate mix on DM basis) or the diet partially replacing barley silage with DDGS and alfalfa hay (DG+AH: 20% barley silage, 20% DDGS, 10% alfalfa hay, and 50% concentrate mix on DM basis). All diets were formulated to contain 19.7% crude protein by replacing beet pulp in DG and DG+AH with corn gluten meal and urea in CON. Compared to CON, DG and DG+AH increased dry matter intake (23.2, 22.7 vs. 20.1 kg/d, $P < 0.0001$), milk yield (26.7, 27.5 vs. 23.9 kg/d, $P < 0.0001$), milk protein yield (0.97, 0.99 vs. 0.87 kg/d, $P < 0.0001$) and body weight gain (385, 408 vs. 84.7 g/d, $P < 0.0001$) but no differences were observed between DG and DG+AH. While milk fat concentration differed ($P < 0.0001$) among the three diets, (3.91%, 3.60% and 3.37% for CON, DG, and DG+AH, respectively), milk fat yield was not affected by treatment ($P = 0.52$) with an average of 0.92 kg/d. The DG and DG+AH decreased chewing time (703, 709 vs. 763 min/d, $P < 0.0001$) and mean ruminal pH (5.89, 5.85 vs. 6.11, $P = 0.005$) and increased the duration that ruminal pH was below 5.8 (667, 705 vs. 438 min/d, $P = 0.02$) with no differences between DG and DG+AH. These results indicate that partially replacing barley silage with DDGS can improve productivity of lactating dairy cows, but it may also decrease chewing time, rumen pH, and milk fat concentration. The dietary inclusion of alfalfa hay may not help alleviate such decreases.

Key Words: barley silage, DDGS, alfalfa hay

Growth and Development

T108 Genetic group and slaughter weight influence on carcass quantitative traits of feedlot cattle. R. Mello*[1], F. D. de Resende[2], A. C. de Queiroz[3], M. H. de Faria[2], P. V. R. Paulino[3], and G. R. Siqueira[2], [1]*Universidade Federal de Roraima*, *Boa Vista, Roraima, Brazil*, [2]*Agência Paulista de Tecnologia dos Agronegócios*, *Colina, São Paulo, Brazil*, [3]*Universidade Federal de Viçosa*, *Viçosa, Minas Gerais, Brazil*.

The purpose of this study was to investigate the genetic group and slaughter weight influence on carcass quantitative characteristics of the cattle. Thirty six young (20 mo) bulls, 18 crossbred F1 Red Angus × Nellore (1/2 RA 1/2 N) and 18 F1 Blonde D'Aquitaine × Nellore (1/2 BA 1/2 N) were used. The young bulls were finished in a feedlot and slaughtered at 480, 520 and 560 kg of shrunk body weight (SBW). A completely randomized experimental design of a 2 × 3 (2 genetic groups × 3 slaughter weights) factorial arrangement with six replicates was used. The animals were slaughtered in a commercial slaughter-house. Data were analyzed with SAS® software using initial SBW as a covariate. The table below shows the least-square means of cold carcass wt (CCW), carcass compactness index (CCI), daily carcass gain (DCG), kidney, pelvic and inguinal fat (KPI), backfat thickness (BFT), ribeye area (REA) and dressing percentage (DP). There was detected effect ($P<0.05$) of genetic group (GG) and slaughter weight (SW) on carcass quantitative characteristics. However, the interaction between GG and SW were not significant ($P>0.05$) for all measured traits. The 1/2 BA 1/2 N young bulls had a higher DCG, absolute (kg) and relative (kg/100kgCCW) KPI than 1/2 RA 1/2 N young bulls, or vice-versa. As the slaughter weight rised the CCW, CCI, absolute (kg) and relative (kg/100kgCCW) KPI, absolute (mm) and relative (mm/100kgCCW) BFT, absolute REA (cm^2) and DP increased; while the DCG decreased with increasing in SW. The relative REA was not affected by the different treatments (28.7 cm^2/100kgCCW). Thereby, finishing of crossbred F1 Blond D'Aquitaine × Nellore young bulls on feedlot until the animals achieve heavier slaughter weights allow the production of the better carcasses.

Table 1. Least square means

	Genetic Group (GG)		Slaughter Weight (SW)		
	½ RA ½ N	½ BA ½ N	480	520	560
CCW, kg	275.9	281.7	250.4^{c}	273.8^{b}	312.2^{a}
CCI, kg/cm	2.1	2.1	1.9^{c}	2.1^{b}	2.3^{a}
DCG, kg/d	1.2^{B}	1.4^{A}	1.5^{a}	1.1^{b}	1.2^{b}
KPI, kg	5.8^{B}	7.6^{A}	4.4^{c}	6.9^{b}	8.9^{a}
KPI, kg/100kgCCW	2.1^{B}	2.7^{A}	1.8^{b}	2.5^{a}	2.9^{a}
BFT, mm	2.9	3.2	2.1^{c}	2.7^{b}	4.4^{a}
BFT, mm/100kgCCW	1.0	1.1	0.8^{b}	1.0^{b}	1.4^{a}
REA, cm^2	80.2	78.0	72.9^{b}	79.2^{ab}	85.2^{a}
DP, %	53.0	53.7	52.3^{b}	52.7^{b}	55.1^{a}

Within a row, means followed by different capital and small letters differ (P<.05), respectively, among GG and SW by Tukey test.

Key Words: beef cattle, carcass dressing, subcutaneous fat thickness

T109 Physical carcass composition of crossbreed beef cattle slaughtered at different end points. R. Mello*[1], F. D. de Resende[2], A. C. de Queiroz[3], M. H. de Faria[2], G. F. Alleoni[2], and P. V. R. Paulino[3], [1]*Universidade Federal de Roraima*, *Boa Vista, Roraima, Brazil*, [2]*Agência Paulista de Tecnologia dos Agronegócios*, *Colina, São Paulo, Brazil*, [3]*Universidade Federal de Viçosa*, *Viçosa, Minas Gerais, Brazil*.

The objective in this trial was to assess the physical carcass composition of finished crossbred feedlot beef bulls and slaughtered at different body masses. Thirty six young (20 mo) bulls, 18 crossbred F1 Red Angus × Nellore (1/2 RA 1/2 N) and 18 F1 Blonde D'Aquitaine × Nellore (1/2 BA 1/2 N) were used. The young bulls were finished on feedlot and slaughtered at 480, 520 and 560 kg of shrunk body weight (SBW). A completely randomized experimental design in a 2 × 3 (2 genetic groups × 3 slaughter weights) factorial arrangement with six replicates was used. Physical composition was predicted for each carcass using separation (bone, lean, fat) of 9-10-11th rib sections from one side. Data were analyzed with SAS® software using initial SBW as a covariate. The table below shows the least-square means of physical carcass composition. There was significant effect ($P<0.05$) of genetic group (GG) and slaughter weight (SW) on physical composition of the carcasses. The interaction between GG and SW were not significant ($P>0.05$) for all measured traits. The 1/2 BA 1/2 N young bulls had a higher ($P<0.05$) lean proportion, lean to bone ratio, and lower ($P<0.05$) bone proportion than 1/2 RA 1/2 N young bulls. The lighter young bulls were associated ($P<0.05$) to a higher proportion of bone, leaner carcass, and lower lean

to bone and fat to bone ratios. The crossbred F1 Blonde D'Aquitaine × Nellore young bulls and heavier animals produced better carcasses in the finishing phase on feedlot than F1 Red Angus × Nellore and lighter animals.

Table 1. Least square means

	Genetic Group (GG)		Slaughter Weight (SW)		
	½ RA ½ N	½ BA ½ N	480	520	560
Bone, %	16.2^{A}	15.3^{B}	17.1^{a}	14.9^{b}	15.3^{b}
Lean, %	61.4^{B}	62.9^{A}	62.3^{ab}	63.2^{a}	61.0^{b}
Fat, %	22.4	21.8	20.6^{b}	21.9^{ab}	23.7^{a}
Lean:Bone ratio	3.8^{B}	4.1^{A}	3.7^{b}	4.3^{a}	4.0^{ab}
Fat:Bone ratio	1.4	1.4	1.2^{b}	1.5^{a}	1.6^{a}

Means followed by different capital and small letters within a row differ (P<0.05), respectively, among GG and SW by Tukey test.

Key Words: breeds, feedlot, 9-11th rib section

T110 Chemical composition of HH section from crossbred beef bulls slaughtered at different body masses. R. Mello*[1], A. C. de Queiroz[2], F. D. de Resende[3], M. H. de Faria[3], G. R. Siqueira[3], and G. F. Alleoni[3], [1]*Universidade Federal de Roraima, Boa Vista, Roraima, Brazil*, [2]*Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil*, [3]*Agência Paulista de Tecnologia dos Agronegócios, Colina, São Paulo, Brazil*.

The aim of the present experiment was to study the effects of genetic groups and slaughter end points on chemical composition of the 9-10-11th rib section from one side to each carcass. Thirty six young (20 mo) bulls, 18 crossbred F1 Red Angus × Nellore (½ RA ½ N) and 18 F1 Blonde D'Aquitaine × Nellore (½ BA ½ N) were used. The young bulls were feedlot finished and slaughtered at 480, 520 and 560 kg of shrunk body weight (SBW). A completely randomized experimental design of a 2 × 3 (2 genetic groups × 3 slaughter weights) factorial arrangement with six replicates was used. Data were analyzed with SAS® software using initial SBW as a covariate. The table below shows the least-square means of dependent variables. The genetic group (GG) and its interaction with slaughter weight (SW) were not significant (P>0.05) for all measured traits. There was significant effect (P<0.05) of slaughter weight on chemical composition of the HH sections. The heavier animals had been lower water content and fatter HH sections than lighter animals at slaughter. The protein and ash contents didn't differ (P>0.05) among the different treatments. Thus, young bulls with heavier slaughter weight have better chemical composition of the HH section than young bulls with heavier slaughter weight, independently of genetic group.

Table 1. Least square means

	Genetic Group (GG)		Slaughter Weight (SW)		
	½ RA ½ N	½ BA ½ N	480	520	560
Water, %	61.0	61.6	63.5^{a}	61.3^{b}	59.1^{c}
Ash, %	6.3	5.7	6.0	6.0	5.9
Protein, %	17.2	17.0	17.3	17.1	17.1
Lipid, %	15.5	15.7	13.2^{c}	15.6^{b}	17.9^{a}

Means followed by different capital and small letters within a row differ (P<0.05), respectively, among GG and SW by Tukey test.

Key Words: breeds, feedlot, 9-11th rib section

T111 Measurement of changes in body composition of piglets from birth to 4 kg using quantitative magnetic resonance (QMR). A. D. Mitchell*[1], G. Taicher[2], and I. Kovner[2], [1]*USDA, Agricultural Research Service, Beltsville, MD*, [2]*Echo Medical Systems, Houston, TX*.

During studies of the growth of neonatal piglets it is important to be able to accurately assess changes in body composition. Previous studies have demonstrated that QMR provides accurate measurements of total body fat, lean, and water in non-anesthetized piglets. The purpose of this study was to use QMR to measure changes in the body composition of piglets during growth from birth to 4 kg BW. Using a QMR instrument (EchoMRI), a total of 60 pigs were scanned an average of 5 times starting at 2.7±1.3 d of age (1.95±0.42 kg) and finally at 13.1±4.3 d (4.14±0.52 kg). Each scan consisted of triplicate measurements. The rates of total body growth and fat and lean deposition were analyzed by linear regression analysis. The mean (±SD) rate of total body growth was 236±76 g/d (R^2=0.98±0.04). The rate of fat deposition ranged from 10.6 to 64.9 g/d with a mean of 32±13 g/d (R^2=0.97±0.04). The rate of lean deposition ranged from 39.1 to 353.6 g/d with a mean of 188±60 g/d (R^2=0.95±0.10). The rates of both fat and lean deposition were highly correlated (P<0.001) with total body growth rate (r = 0.88 and 0.94, respectively). The correlation between the rates of fat and lean deposition was 0.74 (P<0.001). The results of this study demonstrate that QMR is a useful method for measuring changes in body composition in neonatal pigs. Furthermore, the results indicate that during the period of growth from birth to 4 kg, the rates of both fat and lean deposition are linear and highly correlated with total body growth.

Key Words: quantitative magnetic resonance, piglets, body composition

T112 An *in vivo* and *in vitro* comparison of muscle precursor cells originating from broiler and layer chick somites. P. E. Mozdziak*, D. Hodgson, and J. N. Petitte, *North Carolina State University, Raleigh*.

Unique avian genetic resources at NC State University were employed to test the hypothesis that the embryonic environment has a greater impact on muscle precursor cell contribution to growth than the genetically-determined growth potential of the muscle-precursor cells. The somites (#17-20; including a portion of the associated neural tube)that form the right *Pectoralis thoracicus* were removed from broiler chick embryos, which are genetically pre-disposed to form large muscles, and replaced with somites from transgenic, layer strain chick embryos (eGFP or lacZ), which are genetically pre-disposed to form small muscles. Embryos were cultured through hatching, and chicks were grown to 8 weeks of age. Muscle weights were lower ($P < 0.05$) in the transgenic layer-derived muscle (233 g) than the broiler-derived background muscle (244 g). Muscle precursor cell mitotic activity assessed using 5-Bromo-2' deoxyuridine (BrdU) labeling revealed a higher ($P < 0.05$)labeling index in the layer-derived muscle at 8 weeks of age (1.72±0.20) than in the broiler background muscle (0.40±0.01) suggesting that the muscle precursor cells in the layer derived muscle responded to the environmental cues provided by the broiler host, and it also suggests that the layer muscle may contain a smaller complement of muscle precursor cells to support growth. Furthermore, muscle precursor cells were fractionated from the layer-derived and the contra-lateral broiler-derived muscles and inoculated into culture. Muscle precursor cells originating from the layer-derived somites exhibited slower ($P < 0.05$) *in vitro* growth than muscle precursor cells originating from broiler-derived somites suggesting that the muscle precursor cells retained their genetically pre-determined responsiveness to cell culture conditions. The overall results of the study suggest that layer-derived muscle precursor cells

retain their genetic low responsiveness to cell culture conditions, but their proliferative activity *in vivo* reflects a response to environmental cues to support the growth potential reflective of the host animal.

Key Words: avian, embryo, development

T113 Glucose metabolism in preterm (PT) and term (T) born neonatal calves. H. M. Hammon*[1], J. Steinhoff[1], S. Görs[1], C. C. Metges[1], and R. M. Bruckmaier[2], [1]*Institute for the Biology of Farm Animals (FBN), Dummerstorf, Germany*, [2]*University of Bern*, *Bern, Switzerland.*

In neonatal calves, the glucose status often is impaired, even more so in PT born calves. The objective of the present study was to investigate gluconeogenesis (GNG) *in vivo* and hepatic levels of key-enzymes involved in GNG in calves spontaneously born at term (T; n=7) and in calves delivered by sectio 9 d before term (PT; n=7). Calves were gavaged with Deuterium-labeled water (2 × 10 g D_2O /kg BW within 4 h) and received [$U^{13}C$]glucose i.v. (prime: 4.3 μmol/kg BW; infusion: 6.4 μmol/[kg BW × h] for 4 h) after birth. Calves were not fed on d 1 and received milk in amounts of 4% of BW on d 2. Blood samples were taken during tracer administration on d 1 and before and 2 h after milk intake on d 2 to measure total glucose production (GP) and GNG (on d 1) as well as plasma concentrations of glucose photometrically and insulin and glucagon by RIA. Thereafter, calves were slaughtered and liver samples were collected to measure glycogen content and mRNA levels and enzyme activities of pyruvate carboxylase (PC, EC 6.4.1.1), cytosolic phosphoenolpyruvate carboxykinase (PEPCK; EC 4.1.1.32) and glucose-6-phosphatase (G6-Pase; EC 3.1.3.9). Blood data were analyzed by Mixed Model of SAS and GP, GNG, and liver measurements by GLM. Plasma glucose concentrations decreased ($P < 0.05$) from d 1 to d 2 in PT. Plasma insulin concentrations decreased ($P < 0.05$) from d 1 to d 2 and increased ($P < 0.05$) after feed intake on d 2 in both groups. Plasma glucagon concentrations were higher ($P < 0.01$) in PT than T on d 1 and 2. GP and GNG ($P < 0.001$) on d 1 and hepatic glycogen content ($P < 0.05$) on d 2 were higher in T than PT. Enzyme activities of PEPCK ($P < 0.05$) and mRNA levels of PC ($P < 0.1$) were higher in T than PT, but activities of G6-Pase were higher ($P < 0.05$) in PT than T. In conclusion, PT calves showed lower endogenous GP and had difficulty maintaining glucose homeostasis. These data indicated function, but less maturation of hepatic glucose production in PT than T calves.

Key Words: neonatal calf, glucose metabolism, preterm

T114 Milk diet affects glucose status and postprandial hepatic glucose metabolism in neonatal calves. J. Steinhoff*[1], S. Görs[1], C. C. Metges[1], R. M. Bruckmaier[2], and H. M. Hammon[1], [1]*Institute for the Biology of Farm Animals (FBN), Dummerstorf, Germany*, [2]*University of Bern*, *Bern, Switzerland.*

Calves show hypoglycemia at birth and lactose intake does not meet glucose demands. Therefore, we have investigated postnatal endogenous glucose production, especially gluconeogenesis (GNG), and hepatic glucose metabolism in neonatal calves depending on milk intake. Calves were fed twice daily either colostrum (C) or a milk-based formula (F; n=7 per group) with same nutrient density as colostrum, but no biologically active factors. Amounts (per meal) fed were 4% of BW on d 1 and 5% of BW on d 2-4. On d 3, calves went 15 h without food and were then gavaged with Deuterium-labeled water (2 × 10 g D_2O/kg BW within 4 h) and received [$U^{13}C$]glucose i.v. (prime: 4.3 μmol/kg BW; infusion: 6.4 μmol/[kg BW × h] for 4 h) to measure GNG. Blood samples were taken on d 1 before feeding, during tracer administration on d 3 and before and 2 h after feeding on d 4 to measure GNG (d 3) as well as plasma concentrations of glucose, insulin and glucagon. Calves were slaughtered 2 h after feed intake on d 4 and glycogen content as well as mRNA levels and enzyme activities of pyruvate carboxylase (PC, EC 6.4.1.1) and cytosolic phosphoenolpyruvate carboxykinase (PEPCK; EC 4.1.1.32) were measured in liver. Blood data were analyzed by Mixed Model of SAS and GNG and liver measurements by GLM. Plasma glucose and glucagon concentrations increased from d 1 to d 4 in both groups and plasma glucose was higher ($P < 0.05$), but plasma glucagon was lower ($P < 0.001$) in C than F. On d 4, plasma glucose and insulin concentrations increased ($P < 0.05$) after feed intake much more in C than F. GNG on d 3 was not affected by diet. Hepatic glycogen content was higher ($P < 0.001$) in C than F, whereas mRNA levels ($P = 0.1$) and activities ($P < 0.001$) of PC were lower in C than F. In conclusion, elevated plasma glucose concentrations and hepatic glycogen content in C-fed calves were associated with differences in postnatal endocrine changes and hepatic PC activities between C- and F-fed calves, but not with differences in GNG during feed restriction.

Key Words: neonatal calf, colostrum, glucose metabolism

T115 Metabolic maturity at birth and neonate lamb survival and growth. III. Association among pre-suckling plasma metabolic and endocrine factors and lamb growth to weaning. D. R. Miller*[1], R. B. Jackson[1], D. Blache[2], and J. R. Roche[1], [1]*Tasmanian Institute of Agricultural Research*, *Mt Pleasant, TAS, Australia*, [2]*University of Western Australia*, *Perth, WA, Australia*.

This study investigated the metabolic and hormonal characteristics of Dorset cross lambs at birth and their relationship to subsequent growth to weaning. Multiparous, fine-wool Merino ewes (60 ± 6.5 kg BW, n = 150) of equal numbers of single and twin-lamb bearing status were untreated, or treated with 1.5 or 3 mg dexamethasone (DEX) at either Day 130 or 141 of gestation (n = 30 ewes per treatment) and lambed over 20 d on ryegrass dominant pastures. A 4 to 5 ml blood sample was collected from lambs prior to suckling (30 min after birth) and plasma analyzed for glucose, BHBA, urea, NEFA, insulin, ghrelin and leptin. Ewes and lambs grazed perennial ryegrass-based pastures and lamb BW (22.2 ± 3.22 kg), crown to rump length, and heart girth were measured 75 d after the commencement of lambing, which was 3 d before weaning. Lambs (n= 130) grazed together for another 69 d on ryegrass pastures, and then for 4 d on a dual-purpose oat crop before final weighing and condition scoring. Data were analysed using stepwise multiple regression analysis where non-significant ($P > 0.05$) independent variables were removed, with final models including DEX treatment, rate and timing of DEX treatment, and their interaction term. After accounting for DEX effects, pre-suckling plasma leptin levels were negatively associated with rate of weight gain to ($P < 0.05$) and BW at ($P < 0.05$) weaning, and BCS at final weighing ($P < 0.01$). Lambs with greater heart girth at birth were heavier and larger at weaning ($P < 0.01$). Pre-suckling plasma glucose, BHBA, urea, NEFA, insulin, or ghrelin levels were not ($P > 0.05$) associated with growth to weaning or BW at trial completion. In conclusion, these results indicate that birth size and pre-sucking leptin concentrations are associated with subsequent growth rate in lambs.

Key Words: growth, leptin, sheep

T116 Glucagon-like peptide-2 increases splanchnic blood flow acutely in calves but loses effectiveness with chronic exposure. C. C. Taylor-Edwards*[1], D. G. Burrin[2], J. J. Holst[3], K. R. McLeod[1], and D. L. Harmon[1], [1]*University of Kentucky, Lexington*, [2]*USDA/ARS Children's Nutrition Research Center, Baylor College of Medicine, Houston, TX*, [3]*The Panum Institute, University of Copenhagen, Copenhagen, Denmark*.

Glucagon-like peptide-2 (GLP-2) is a 33-amino acid hormone secreted from the gastrointestinal tract that rapidly increases small intestinal blood flow. No experiments have been conducted evaluating the blood flow response to GLP-2 after extended administration, nor have investigations been performed in ruminants. Eight Holstein calves with an ultrasonic flow probe around the superior mesenteric artery (SMA) and catheters in the carotid artery, mesenteric vein, portal vein, and hepatic vein were paired by age and randomly assigned to treatment: Control (0.5% BSA in saline; n=4) or GLP-2 (50 μg/kg BW bovine GLP-2 in vehicle; n=4). Treatments were administered by subcutaneous injection every 12 h for 10 d. Calves were fed a 50:50 (DM basis) mixture of alfalfa cubes and calf starter at 2.75% of BW in 2 daily meals. A blood flow experiment was conducted on d 0 (Acute) and d 10 (Chronic) of administration and consisted of 3 periods: baseline saline infusion (30 min) to establish baseline blood flow, treatment infusion in which calves were infused with their assigned treatment, either BSA or GLP-2 (1000 pmol/kg/h) for 60 min, and saline infusion (60 min) to observe the recovery of blood flow after treatment infusion. Portal and hepatic plasma flows were measured by *p*-aminohippurate dilution. The statistical model included infusion, treatment, and their interaction as fixed effects and block as a random effect. Repeated measures were conducted on infusion with calf(trt) as the subject. Infusion of GLP-2 increased SMA blood flow to 175% of baseline on d 0 but to only 137% of baseline after Chronic treatment (interaction, P=0.0002). Similar trends were observed for portal and hepatic plasma flow. Our results show that GLP-2 increases splanchnic blood flow in ruminants but this response is attenuated after 10-d GLP-2 administration. These results suggest that GLP-2 could modulate nutrient absorption in ruminants through effects on splanchnic blood flow.

Key Words: glucagon-like peptide-2, gastrointestinal growth, hormone

T117 Glucagon-like peptide-2 increases small intestinal mass of calves. C. C. Taylor-Edwards*[1], D. G. Burrin[2], K. R. McLeod[1], and D. L. Harmon[1], [1]*University of Kentucky, Lexington*, [2]*USDA/ARS Children's Nutrition Research Center, Baylor College of Medicine, Houston, TX*.

Glucagon-like peptide-2 (GLP-2) is a 33-amino acid hormone secreted from the gastrointestinal tract in response to luminal nutrients that potently increases small intestinal mass in non-ruminants. However, the effects of GLP-2 on small intestinal mass and morphology of ruminants is unknown. Eight Holstein calves were paired by age and randomly assigned to treatment: Control (0.5% BSA in saline; n=4) or GLP-2 (50 μg/kg BW bovine GLP-2 in vehicle; n=4). Treatments were administered by subcutaneous injection every 12 h for 10 d. Calves were fed a 50:50 (DM basis) mixture of alfalfa cubes and calf starter at 2.75% of BW in 2 daily meals. On d 11, 2 h after an intravenous injection of 10 mg/kg BW 5-bromo-2′-deoxyuridine (BrdU), calves were killed, gastrointestinal tissues weighed, and epithelial samples obtained from the rumen, omasum, abomasum, duodenum, jejunum, ileum, and colon. Samples were analyzed for villus height, crypt depth, and BrdU staining. The statistical model included treatment as a fixed effect and block and block by treatment as random effects. Compared to Control, GLP-2 increased small intestinal mass by 24% (P=0.04). Intestinal length was unchanged but intestinal thickness was increased (P=0.02) by GLP-2, particularly because of increased epithelial mass in the jejunum (P=0.008) and ileum (P=0.04). Treatment with GLP-2 increased villus height in the duodenum (P=0.03), jejunum (P=0.06), and ileum (P=0.09). In addition, GLP-2 increased crypt depth in the duodenum (P=0.06) and jejunum (P=0.02). Treatment with GLP-2 increased crypt cell proliferation as indicated by increased BrdU-labeling in the duodenum (P=0.02), jejunum (P=0.01), and ileum (P=0.05). These results demonstrate that GLP-2 induces similar increases in ruminant intestinal growth as observed in non-ruminants and could be an important mediator of small intestinal growth in response to luminal nutrients in ruminants.

Key Words: glucagon-like peptide-2, gastrointestinal growth, hormone

T118 Maternal low and high protein diets during pregnancy affect body weight and stress reactivity in the offspring of pigs. M. Graebner*, E. Kanitz, M. Tuchscherer, B. Stabenow, C. C. Metges, C. Rehfeldt, and W. Otten, *Research Institute for the Biology of Farm Animals (FBN), Dummerstorf, Germany*.

Objective: Nutritional imbalance during pregnancy can have long-term consequences on the development of the offspring. An inadequate nutrient supply and/or maternal hormones can cause fetal growth retardation, metabolic changes and alterations of stress-sensitive systems in the offspring. In this study, we determined the effects of low and high protein diets in pregnant sows on growth development and function of the HPA and SAM axis of their offspring. Methods: Forty-two German Landrace sows (first parity) were fed isocaloric diets with high (HP, 30%), low (LP, 6%) or control (CP, 12%) protein levels throughout gestation. The offspring was reared by foster sows until weaning on postnatal day (PND) 28. Salivary cortisol was measured in sows during gestation, and plasma cortisol and catecholamines were determined in the offspring on PND 1 and 27 (basal levels) as well as under challenging conditions (weaning, LPS-, ACTH- and insulin-challenge) later in life. Glucocorticoid receptor (GR) binding and neurotransmitter concentrations were measured in stress-related brain areas. Results: The LP diet caused a decreased growth performance of sows and reduced the birth weights of piglets. Salivary cortisol levels were increased in LP sows from mid gestation until parturition. HP piglets showed a decreased serotonergic activity in the locus coeruleus on PND 1. The GR binding in the hippocampus was increased in LP and HP offspring on PND 27. Basal stress hormone concentrations were not affected, however, there was a tendency for a higher cortisol response to weaning in female LP piglets. In addition, LP offspring showed an increased catecholamine response during an insulin challenge test. Conclusion: The present results indicate that dietary protein levels during pregnancy in sows can influence growth and the function of stress systems in the offspring. In particular, a dietary protein deficiency resulted in growth retardation in the offspring and an increased reactivity of their SAM system.

Key Words: fetal programming, stress reactivity, glucocorticoids

T119 Linoleic acid changes fatty acid profiles and alters gene expression in bovine adipocyte cultures. A. P. Burns*, S. K. Duckett, S. L. Pratt, and S. E. Ellis, *Clemson University, Clemson, SC*.

The objective of this study was to determine if differences in fatty acid profiles or gene expression exist when adipocytes are exposed to a linoleic acid (C18:2)-supplemented media post differentiation. With

limited information in the literature about the timing of lipid uptake in differentiated preadipocytes, a secondary objective of this study was to evaluate fatty acid composition and gene expression over time. Primary preadipocyte cultures were isolated from 18 mo old Angus crossbred steers. After plated cells reached confluence (d 0), differentiation media was applied for 2 d and cells were supplemented with 0 mM (C) or 0.3 mM (LA) C18:2 for 10 d. Cells were harvested on d 2, 6, and 12 for analysis using gas chromatography and RT-PCR. The percentages of palmitic (C16:0), stearic (C18:0), oleic (C18:1), and linolenic (C18:3) acids were decreased (P<0.001) in LA cells compared to C cells on d 6 and 12. LA cells had greater (P<0.001) C18:2 and total fatty acid content compared to C, in terms of percent composition and μg/well. Analysis of mRNA expression revealed that fatty acid synthase (FASN) and glucose transporter 4 (GLUT4) mRNAs were elevated (P<0.05) in LA cells compared to C. Regardless of C18:2 supplementation, FASN mRNA levels increased (P<0.001) and stearyl-CoA mRNA levels decreased (P<0.0001) over time. A significant interaction existed between time and treatment for sterol regulatory element binding protein, peroxisome proliferator-activated receptor gamma, and adipocyte protein 2 mRNA levels. C cells decreased (P<0.05) in their expression of all these mRNAs over time, whereas LA cells increased (P<0.05) slightly or stayed the same. In conclusion, supplementing culture media with C18:2 produced changes in fatty acid composition of bovine adipocytes by d 6 and influenced mRNA expression. Based on our data of fatty acid uptake and mRNA expression changes of differentiation-associated proteins, we propose to time future investigations of bovine adipocytes more closely with differentiation.

Key Words: adipocyte, linoleic acid, gene expression

T120 Docosahexaenoic acid enhances hepatic serum amyloid A expression via a protein kinase A-dependent mechanism. J. J. Dai, Y. C. Wang, P. H. Wang, H. J. Mersmann, and S. T. Ding*, *Institute of Biotechnology, National Taiwan University, Taipei, Taiwan*.

Serum amyloid A (SAA) can induce adipocyte lipolysis to reduce fat deposition in adipocytes and hepatocytes. Hepatic SAA1 mRNA is increased by docosahexaenoic acid (DHA) treatment in pigs. Understanding whether DHA can decrease fat deposition through SAA1 and what the mechanism is would help to develop strategies against the fatty liver symptom and obesity. In the current study, we demonstrated that DHA treatment simultaneously increased human SAA1 and C/EBP β mRNA expression in human hepatoma cells, SK-HEP-1. Utilizing a promoter deletion assay, we found that a C/EBPβ binding site in the SAA1 promoter region between -242 and -102 bp was crucial for DHA-mediated SAA1 expression. When we mutated the putative C/EBPβ binding site, the DHA-induced increment of SAA1 promoter activity was suppressed, suggesting that this binding sequence is very important for the DHA regulated function. The DHA also increased C/EBP β protein expression and the addition of a protein kinase A inhibitor, H89, abrogated the increase in C/EBPβ. In addition, the up-regulation of SAA1 expression by DHA was inhibited by H89. These data suggest that DHA up-regulates the expression of C/EBPβ by activating PKA. Indeed, DHA treatment increased both PKA activity and the expression of C/EBPβ in SK-HEP-1 cells. We have demonstrated that DHA activates PKA to increase the expression of SAA1. Because the increase of SAA1 will increase the lipolytic activity by increasing the expression of lipolytic genes, such as hormone sensitive lipase, DHA could reduce lipid accumulation in the liver and the body. These results may be useful to develop new approaches to reduce body fat deposition and fatty liver.

Key Words: DHA, SAA, C/EBPbeta

T121 Effects of arginine supplementation to gilts during early gestation on fetal myogenesis. C. Kalbe*[1], M. Porm[1], J. Bérard[2], G. Bee[2], and C. Rehfeldt[1], [1]*Research Institute for the Biology of Farm Animals, Dummerstorf, Germany*, [2]*Agroscope, Liebefeld Posieux, Switzerland*.

To study the impact of additional L-arginine intake during early gestation on cellular properties of skeletal muscle of the offspring, 20 Swiss Large White gilts were randomly allocated to either control (C) or arginine (A) group at the day of mating. Gilts were fed 3 kg daily of a standard diet and A-gilts received additionally 26 g/d L-arginine from d 14 to 28 of gestation. At d 75 of gestation the gilts were sacrificed with 3 C-gilts being excluded because of non-pregnancy. From each litter the lightest, heaviest, and one fetus with an average fetal weight from both genders were selected and the *longissimus* muscle (LM) was excised for analyses (n = 98 in total). Compared with the offspring from the C-gilts the protein/DNA ratio tended to be decreased (32.5 vs. 34.2; P = 0.08) in response to maternal arginine supplementation. This resulted from a numeric increase (1.80 vs. 1.74 mg/g tissue; P = 0.16) in DNA concentration and a concomitant numeric decrease in protein concentration (57.91 vs. 59.23 mg/g tissue; P = 0.36). Moreover, treatment × gender interactions revealed lower creatine kinase (CK) activity (P < 0.05) and protein concentration (P = 0.07) in female offspring of arginine-supplemented gilts. The mRNA expression of genes encoding for the muscle regulatory factors (MRF) was measured in the LM of light and heavy female fetuses (n = 28). In offspring of A-gilts the transcript expression of the Myf5 gene was increased (1.41 vs. 1.00; P < 0.05) compared with the offspring of C-gilts. The mRNA expression of the MyoD, myogenin and MRF4 genes remained unchanged by arginine intake. The results suggest that arginine supplementation during early gestation stimulates myogenic proliferation associated with a lower degree of muscular maturity at d 75 of gestation indicative of delayed differentiation. This effect is particularly pronounced in female offspring.

Key Words: pregnant gilts, arginine, myogenesis

T122 Identification and characterization of the bovine G protein-coupled receptor GPR41 and GPR43 genes. A. Wang, Z. Gu, B. Heid, R. M. Akers, and H. Jiang*, *Virginia Polytechnic Institute and State University, Blacksburg*.

Volatile fatty acids (VFA) regulate rumen development, insulin and glucagon secretion, and some other physiological processes in cattle. The underlying mechanisms are unknown. Recent "reverse pharmacology" studies identified human G protein-coupled receptors GPR41 and GPR43 as receptors for short-chain fatty acids. It is possible that proteins similar to human GPR41 and GPR43 mediate the regulatory effects of VFA in cattle. In this study, we determined first whether the bovine genome contains genes similar to the human GPR41 and GPR43 genes and secondly whether and where these genes are expressed in cattle, and if the proteins encoded by these genes can be activated by acetate, propionate, and butyrate. A search of GenBank revealed bovine genomic sequences and expressed sequence tags highly similar to the human GPR41 and GPR43 DNA and cDNA sequences. The protein-coding and 5′ untranslated regions of the bovine GPR41 and GPR43 mRNA were cloned and sequenced from the spleen tissue. Based on these sequences, the bovine GPR41 gene contains three exons and its transcription is initiated at two leader exons, generating two GPR41 mRNA variants differing in the 5′ untranslated region. The bovine GPR43 gene contains two exons, and transcription of this gene is initiated from a single start site. The amino acid sequences deduced from the bovine GPR41 and GPR43 mRNA sequences are more than 75% identical to those of the human GPR41 and GPR43 and are predicted to encode

seven transmembrane domains, typical of G protein-coupled receptors. By RT-PCR, both bovine GPR41 and GPR43 mRNA were detected in a variety to tissues, including rumen and pancreas. In a cell system, interaction of the overexpressed bovine GPR41 or GPR43 protein with acetate, propionate, or butyrate inhibited luciferase reporter expression from a cAMP-responsive promoter, suggesting that the bovine GPR41 and GPR43 proteins couple to Gαi/11. In total these results demonstrate that the bovine genome encodes functional GPR41 and GPR43 genes and suggest that GPR41 and GPR43 may play a role in the regulatory effects of VFA in cattle.

Key Words: G-protein-coupled receptors, cattle, sequence

T123 Potential role of low-density lipoprotein receptor-related protein (LRP)-1 and IGFBP-3 in the proliferation-suppressing actions of TGF-beta on cultured myogenic cells. E. Kamanga-Sollo, M. S. Pampusch, M. E. White*, M. R. Hathaway, and W. R. Dayton, *University of Minnesota*, *St. Paul*.

Myostatin and transforming growth factor (TGF)-beta suppress both proliferation and differentiation of cultured myogenic cells. Recent studies have shown that the IGF-independent actions of insulin-like growth factor binding protein (IGFBP)-3 facilitate the proliferation-suppressing actions of both myostatin and TGF-beta on cultured myogenic cells; however, the mechanism of this facilitation is not known. To assess this mechanism, we have transfected L6 myogenic cells, which do not produce detectable levels of IGFBP-3, with a construct containing the porcine IGFBP-3 cDNA behind a constitutively active promoter. These transfected cells (tL6) constitutively express porcine IGFBP-3. Consistent with our previous observation that IGFBP-3 facilitates the proliferation-suppressing actions of TGF-beta, dose response curves showed that proliferation of tL6 cells was inhibited at lower TGF-beta concentrations than was proliferation of mock-transfected or non-transfected L6 cells ($p < 0.05$). In non-muscle cells, IGFBP-3 suppresses proliferation by binding to LRP-1, suggesting that binding to this receptor may play a role in the ability of IGFBP-3 to facilitate the proliferation-suppressing actions of TGF-beta and myostatin on cultured myogenic cells. To assess the role of LRP-1 in this process, we have examined the effects of Receptor Associated Protein (RAP), a protein which inhibits ligand binding to LRP-1, on TGF-beta-induced suppression of proliferation of mock-transfected L6 and tL6 myogenic cells. RAP significantly ($p < 0.02$) increases proliferation of TGF-beta-treated tL6 cells, which produce IGFBP-3, while having no effect on proliferation of TGF-beta-treated mock transfected or control L6 cells, neither of which produces IGFBP-3. These data suggest that binding of IGFBP-3 to LRP-1 may play a role in the mechanism by which IGFBP-3 facilitates the proliferation-suppressing actions of TGF-beta on cultured myogenic cells.

Key Words: IGFBP-3, TGF-beta, muscle

T124 Clofibrate treatment up-regulates hepatic gene expression encoding fatty acid oxidation and ketogenesis enzymes in liver of pigs during early postnatal development. K. Shim, L. Xi*, S. Jacobi, and J. Odle, *North Carolina State University*, *Raleigh*.

The aim of this research was to investigate the effects of clofibrate on gene expression of hepatic fatty-acid-oxidation and ketogenesis enzymes in pigs during early neonatal development. Sixty colostrum-deprived newborn pigs were fed milk replacer and orally gavaged with either vehicle (2% Tween 80) or clofibrate (75 mg /kg body weight) ± etomoxir (5 mg/ kg body weight) once daily. The effect of treatment was evaluated on days 0, 1, 4 and 7. Clofibrate treatment did not significantly change liver weights. Messenger RNA abundances measured via qRT-PCR, were increased for carnitine palmitoyltransfersae I (CPT I; 2.8 fold), carnitine palmitoyltransfersae II (CPT II; 3.2 fold), and mitochondrial 3-methly-3-hydroxyglutaryl-CoA synthase (mHMG-CoA-S; 3.8 fold) in pigs fed clofibrate compared to vehicle. Addition of etomoxir did not affect mRNA abundances induced by clofibrate. Transcript levels increased as piglets aged, but the abundances remained relatively constant for CPT I and mHMG-CoA-S after four days and for CPT II after one day. There was no interaction between clofibrate treatment and age. Acyl-CoA oxidase, acetyl-CoA carboxylase and peroxisome proliferator-activated receptor α abundance were not altered by clofibrate, etomoxir or piglet age. In conclusion, clofibrate strongly up-regulates genes of fatty acid oxidation in the young, postnatal pig, but induction is not influenced by developmental age. *Supported by CSREES, USDA NRI program award 2007-35206-1 7897.*

Key Words: clofibrate, fatty acid oxidation, gene expression

T125 Use of gas chromatography to measure stearoyl-CoA desaturase activity and substrate preference. J. A. Stamey*, C. A. Umberger, M. D. Hanigan, and B. A. Corl, *Virginia Polytechnic Institute and State University*, *Blacksburg*.

Stearoyl-CoA desaturase (SCD) catalyzes the desaturation of saturated fatty acyl-CoA substrates, primarily synthesizing palmitoleoyl-CoA and oleoyl-CoA. SCD is a key enzyme controlling milk and body fat composition. Two known isoforms, SCD1 and SCD5, are present in the bovine genome, and found in adipose and brain tissue, respectively. Most SCD activity assays use radiolabeled substrates, and the variety of radiolabeled substrates available for assay can be limited. We developed a method permitting use of a variety of unlabeled fatty acid substrates to measure desaturase activity with gas chromatography based on procedure of Su and Brenna (Anal. Biochem. 261:43). Enzyme reactions (500 μL) were incubated at 37°C for 30 min, stopped with 12% ethanolic KOH, and saponified at 70°C for 30 min. Desaturase activity was assayed using myristate, palmitate, deuterium-labeled stearate, and arachidate complexed to bovine serum albumin as substrates. For myristate, palmitate, and arachidate, monounsaturated product fatty acids were undetectable at time 0, so labeled substrate was not required. Total lipids were extracted from enzyme reactions, methylated, and quantified by gas chromatography using nonadecanoic acid as the internal standard. Due to high levels of endogenous oleate in microsomes, stable-isotope-labeled stearate was used as a substrate and quantification of labeled oleate produced by SCD was performed using gas chromatography-electron ionization mass spectrometry. The assay was linear with microsomal protein up to 200 μg and time up to 45 min. Bovine adipose and brain microsomes were isolated from 14-20-mo old Angus and Angus-cross steers (n=3). Substrate-specific activities in adipose microsomes (100-200 μg protein) were 0.99±0.24, 1.02±0.76, 1.17±2.24, and 0.92±0.27 nmol/mg•min for myristate, palmitate, stearate, and arachidate, respectively. Brain microsome SCD activity was not detected. Application of this method will allow tissue-specific SCD isoform characterization, including substrate specificities and enzyme kinetics.

Key Words: desaturase, microsomes, gas chromatography

T126 Maternal weight and P8 fat amount affects *IGF2* expression in *semitendinosus* muscle tissue of the developing fetus. C. J. Fitzsimmons*[1,2], R. Feldmann[1], Z. A. Kruk[1,3], S. Truran[1], D. Lines[1], D. Rutley[1], and S. Hiendleder[1,4], [1]*JS Davies Epigenetics and Genetics Group, Discipline of Agricultural and Animal Science, The University of Adelaide, Roseworthy Campus, Roseworthy, South Australia, Australia*, [2]*Agriculture and Agri-Food Canada, Department of Agricultural, Food, and Nutritional Science, University of Alberta, Edmonton, Alberta, Canada*, [3]*Chungnam National University, Daejeon, South Korea*, [4]*Research Centre for Reproductive Health, The University of Adelaide, Adelaide, South Australia, Australia*.

Insulin-like growth factor 2 (*IGF2*) is an important promoter and regulator of fetal growth. We have measured *IGF2* exon 10 expression (present in almost all *IGF2* transcripts), in *semitendinosus* muscle of bovine fetuses obtained at Day 153 of pregnancy. Relative *IGF2* expression was examined using a univariate GLM including factors dam genetics, sire genetics, fetus sex, and covariate dam weight, and their 2, 3, and 4-way interactions, as explanatory variables in the model. There was a strong negative relationship between dam weight and expression of *IGF2* exon 10 in fetal muscle (P=0.015), and the average *IGF2* expression level of male fetuses was higher than that of females (P=0.025). To investigate the origin of the curious negative relationship between fetal *IGF2* expression and maternal weight, empty uterus weight of the dam was substituted into the model for dam weight. The results showed that uterus weight and fetal sex were both significant (P=0.049 and 0.025, respectively). Therefore, maternal size itself had a great influence on fetal gene expression, although there appeared to be some other, possibly nutritional, components involved. Several other measures of change in dam weight during pregnancy were tested in the model, but were not statistically significant. When P8 fat (fat depth as measured on top of the rump of the carcass) was used in the model instead of dam weight, it also was not significant. However, when both P8 fat and dam weight were included in the model, the R^2 value increased from 0.816 to 0.906. Now fetal sex*dam weight*P8 fat and dam genetics*P8 fat were significant (P=0.011 and 0.002, respectively). It also appeared that there was a particular dam weight where the relationship between an increase in maternal P8 fat and fetal *semintendinosus IGF2* expression changes.

Key Words: IGF2, gene expression, pre-natal

T127 Fetal growth is substantially modulated by at least two different genetic loci in the middle part of bovine chromosome 6. A. Eberlein[1], A. Takasuga[2], K. Setoguchi[3], R. Pfuhl[1], K. Flisikowski[4], R. Fries[4], N. Klopp[5], K. Suhre[5], R. Weikard[1], and Ch. Kühn*[1], [1]*Research Institute for the Biology of Farm Animals, Dummerstorf, Germany*, [2]*Shirikawa Institute of Animal Genetics, Fukushima, Japan*, [3]*Cattle Breeding Development Institute of Kagoshima Prefecture, Kagoshima, Japan*, [4]*Chair of Animal Breeding, Technische Universität München, Freising, Germany*, [5]*Helmholtz Zentrum, Munich, Germany*.

In cattle, fetal growth has raised attention for its impact on stillbirth and dystocia in conventional reproduction and also due to the still unknown fetal growth mechanisms impairing calves generated by somatic nuclear transfer techniques. For our analyses of the genetic background of fetal growth, birth weight data were available from a F_1 and F_2 resource population generated from purebred Charolais and German Holsteins by consistent application of embryo transfer. This approach enabled the specific analysis of the genetic background for divergent fetal growth dissected from maternal effects. After genotyping of 16 microsatellite markers, an initial line cross regression model assuming alternatively fixed QTL alleles in the founder breeds revealed a genome-wide significant QTL (F = 90.16, p<0.0001) in the middle part of BTA6. The QTL accounted for 16.3% of the genetic variance in the model and was located within a confidence interval of 12 cM. To further refine the QTL additional 36 targeted SNP were genotyped. Consecutive variance component QTL analyses fitting the QTL and an infinitesimal animal effect revealed that the QTL initially appearing as one, could be divided into two separate loci: one of the QTL, located at 49 cM (LRT 1 QTL vs. no QTL = 94.08), showed a fixed QTL effect. In contrast, the second QTL at 55 cM (LRT 2 QTL vs. 1 QTL = 70.45) evidently had a random effect without fixation of alleles in the founder breeds. While the QTL seem to have a major effect on intrauterine growth, no effect could be detected for the consecutive period from birth to day 121. Association analyses ($p<10^{-15}$) in line with concordance analyses of significantly segregating F_1 individuals identified a SNP at 49 cM, which is in very close linkage disequilibrium with a causal mutation affecting fetal growth or might be a causal mutation itself. The substantial effect size of the detected genetic locus will facilitate further functional investigations of its role in affecting growth during normal and aberrant fetal development in conventional reproduction or in assisted reproduction techniques.

Key Words: fetal growth, cattle, QTL

T128 Relationships between growth and metabolic parameters of replacement heifers on two nutrition programs. F. Abeni[1], L. Calamari[2], G. Pirlo*[1], and L. Stefanini[3], [1]*CRA-FLC, Cremona,Italy*, [2]*Istituto di Zootecnica, U.C.S.C., Piacenza, Italy*, [3]*Azienda Sperimentale V. Tadini, Gariga di Podenzano, Italy*.

In two experimental herds 141 replacement heifers were reared with the aim of investigating the relationships between growth from 5 to 18 mo of age and metabolic parameters. The heifers of both herds were fed on a moderate ADG (MADG: 0.7 kg/d) or on an accelerated ADG (AADG: 0.9 kg/d) diet. Heifers' BW, BCS, wither height (WH), hip height (HH), body length (BL), and hearth girth (HG) were recorded every 28 days. At 9 and 15 mo of age a blood sample was drawn for analysing metabolic profile. Data from body measurements were processed by analysis of variance and growth curve analysis, using Laird's form of Gompertz curve. Metabolic profiles were processed by Principal Components Analysis (PCA), followed by Pearson's correlation analysis between growth curve coefficients and PCs. The AADG heifers had greater actual ADG (0.83 vs 0.74 kg, P<0.001) and greater daily increase of HG (1.76 vs 1.65 mm, P<0.005) than MADG heifers. From 9 to 18 mo of age, BCS of AADG heifers diet was greater than BCS of MADG heifers (P<0.05). From the comparison of growth curve parameters between ADG groups, only the initial growth rate of HG was affected by diet, and no effect was found on maturation rate. The PCA showed a PC1 mainly composed of total protein and albumin (9 and 15 mo), Na and ceruplasmin (9 mo), glucose and Ca (15 mo). This component was positively correlated (P<0.05) with the maturation rate of the growth functions for BW, WH, HH, and HG.

Table 1. Coefficients of the growth curves

		MADG		AADG		
		Mean	SE	Mean	SE	P
BW	b_1_	2.874	0.0637	3.026	0.0849	0.15
	b_2_	0.0035	0.0001	0.0034	0.0002	ns
WH	b_1_	0.748	0.0199	0.761	0.0255	ns
	b_2_	0.0031	0.0001	0.0032	0.0002	ns
HH	b_1_	0.683	0.017	0.687	0.022	ns
	b_2_	0.0036	0.0002	0.0037	0.0001	ns
BL	b_1_	0.944	0.028	0.943	0.037	ns
	b_2_	0.0039	0.0002	0.0040	0.0002	ns
HG	b_1_	1.027	0.0235	1.113	0.0313	0.03
	b_2_	0.0035	0.0004	0.0042	0.0006	ns

Key Words: replacement heifers, ADG, metabolic profile

T129 Luminal energy supply (but not substrate) affects expression of mRNA for three proteins capable of amino acid transport by ileal epithelium (but not duodenal or jejunal) of forage-fed growing beef cattle. S. F. Liao*, J. A. Boling, and J. C. Matthews, *University of Kentucky*, *Lexington*.

To complete our *in vivo* study of whether basal expression of 20 amino acid (AA) transporter genes by duodenal (D), jejunal (J), and ileal (I) epithelia is affected by increased luminal supply of rumen-derived microbes (hence, AA substrates), starch-derived energy, or both, 18 ruminally and abomasally catheterized Angus steers (BW ~ 260 kg) were assigned (n = 6) to either water (basal) or ruminal or abomasal corn starch hydrolysate (SH, by α-amylase) infusion treatment (at 20% ME intake) and fed an alfalfa-cube based diet at 1.3× NE_m requirement. After a 14- or 16-d infusion, steers were killed, small intestinal epithelia harvested, and total RNA extracted. Real-time RT-PCR analysis of the effect of substrate and energy supply on expression of mRNA encoding AA transport systems x^-_c (xCT), T (TAT1), H^+-dependent AA (PAT1), and H^+-dependent peptide (PepT1) was conducted to quantify the relative expression of AA transporter mRNA:18S rRNA. Basal expression of mRNA differed ($0.01 \leq P \leq 0.09$) among the 3 epithelia. For xCT, TAT1, and PepT1, the expression pattern was D = J > I. For PAT1, however, the pattern was J > D = I. Increased luminal AA supply did not affect transporter mRNA content. Although, D and J expression were unaffected by increased luminal energy supply, I expression of xCT mRNA was increased by 41% ($P = 0.04$), PepT1 by 162% ($P = 0.03$), and TAT1 by 56% ($P = 0.08$). The findings that PAT1, TAT1, and xCT mRNA are expressed by cattle small intestinal epithelia are novel. Given the known localization and function of the proteins encoded by the evaluated mRNA, this study suggests that the potential for I apical uptake of luminal peptide-bound AA by PepT1, basolateral transfer of aromatic AA by TAT1 into blood, and cystine uptake from blood by xCT, is enhanced by increased luminal starch-derived energy supply to forage-fed growing cattle.

Key Words: bovine, nutrient-gene interaction, substrate reguation

T130 Early life management and long term productivity of dairy calves. F. Soberon*, E. Raffrenato, R. W. Everett, and M. E. Van Amburgh, *Cornell University*, *Ithaca, NY*.

For many years, early life management of the calf has focused on survival rates and rumen development. However, recent studies suggest that colostrum status as well as nutritional status during the pre-weaning phase may have long term carry-over effects on milk yield potential. The objective of this study was to investigate this relationship in the Cornell Dairy Herd using a test day model (TDM) to evaluate the lactation response over eight years. The management objectives of the calf program have been to double the birth weight by weaning through increased milk replacer intake. The TDM was utilized to generate lactation residuals accounting for the effects of test day such as calving season, days carried calf, days in milk, and lactation number (Everett and Schmitz, 1994; Van Amburgh et al., 1997). Lactation residuals from the TDM were generated from 792 heifers with completed lactations and linear regressions were run on several measures of pre-weaning growth performance, management factors and TDM milk yield solutions. Significant correlations were found for pre-weaned average daily gain (ADG), weaning weight, year and month of birth. Pre-weaning ADG ranged from 0.13 kg to 1.23 kg and ADG had the greatest correlation with first lactation milk production. Using the TDM solutions, for every 1 kg of pre-weaning ADG, heifers produced 1,067 kg more milk during their first lactation ($P < 0.01$). Further, pre-weaning ADG accounted for 25 percent of the variation in first lactation milk yield. Other factors analyzed included age at first calving and birth weight but correlations with TDM lactation residuals were not significant. Data from other farms have yielded similar results and further work, incorporating multiple farms and data sets, is being conducted to better describe the correlation between early life nutritional status and long term productivity. These results demonstrate that increased growth rate, primarily through increased liquid feed intake, prior to weaning has positive effects on lactation milk yield.

Key Words: calves nutrition, milk production, test day model

T131 Metformin inhibits adipogenesis in fetal muscle of dam receiving high energy diet. J. F. Tong*, X. Yan, J. X. Zhao, and M. Du, *University of Wyoming*, *Laramie*.

At the later stage of fetal skeletal muscle development, adipogenesis within fetal muscle forms intramuscular adipocytes, which provide sites for intramuscular fat accumulation (marbling). High energy diet inhibits AMP-activated protein kinase (AMPK) and promotes adipogenesis in fetal muscle, but the cause and effect relationship between AMPK and adipogenesis in fetal muscle has not been established. The objective of current study is to evaluate the role of AMPK in adipogenesis in fetal skeletal muscle. Prior to pregnancy, female weanling C57BL/6J mice were randomized to receive either a control diet (Con, 10% energy by fat) or a high energy diet (Ob, 45% energy by fat) for 10 weeks. At three months of age, mice were mated. The Con mice continued to be fed with control diet, while the Ob mice were separated into 2 groups, with one group fed with high energy diet and the other group fed high energy diet plus metformin (2 mg/ml in drinking water, Met), a known activator of AMPK. Hindlimb and back muscles from newborn mice (Day 1, n = 6 per treatment) were collected for analyses. AMPK phosphorylation was lower in neonatal Ob skeletal muscle. β-Catenin and myogenic regulatory factors MyoD and myogenin were down-regulataed in Ob muscle, while the adipogenic marker, peroxisome proliferator-activated receptor γ (PPARγ), was up-regulated compared to Con muscle, indicating the down-regulation of myogenesis but up-regulation of adipogenesis in Ob neonatal muscle. Metformin administration stimulated AMPK activity, restored MyoD expression and decreased PPARγ expression. Hematoxylin and eosin staining showed more adipocytes were deposited in Ob neonatal skeletal muscle, but metformin reduced the deposition of intramuscular adipocytes. These data indicate that maternal high energy

diet enhances adipogenesis in fetal muscle via inhibition of AMPK. Therefore, AMPK is an attractive molecular target to enhance adipogenesis in fetal muscle and, thus, marbling in the muscle of offspring.

Key Words: adipogenesis, skeletal muscle, AMP-activated protein kinase

T132 Albumin induced cytokine expression in porcine adipose tissue explants. T. G. Ramsay*, M. Stoll, and T. J. Caperna, *USDA-ARS, Beltsville, MD*.

Albumin has historically been included in medium designed for use with adipose tissue when evaluating metabolism, gene expression or protein secretion. However, recent studies with mouse adipocytes (Ruan et al., J. Biol. Chem. 278:47585-47593, 2003) and human adipose tissue (Schlesinger et al., Amer. J. Physiol. 291:27-33, 2006) have demonstrated an acute cytokine response by cells derived from adipose tissue to albumin. The present study was designed to determine whether or not albumin can alter cytokine expression in porcine adipose tissue explants relative to explants not exposed to albumin. Subcutaneous adipose tissue explants (100 mg) were prepared from dorsal subcutaneous adipose tissue of 21 day old pigs using a Stadie-Riggs microtome. Slices were placed in Hanks′ balanced salt solution prior to albumin exposure. Triplicate slices were frozen in liquid nitrogen (controls). Additional slices were incubated in medium 199 supplemented with 100 mM HEPES and 0.5% bovine albumin (Sigma A7030) at 37°C. Triplicate tissue slices were removed from the medium at 1, 2 or 4 hours following exposure to the albumin containing medium and frozen in liquid nitrogen. Samples were extracted for RNA using Qiagen columns and RNA (1 ug) was reverse transcribed and used in real-time PCR analysis to assess the expression of the adipokines leptin, adiponectin, tumor necrosis factor alpha (TNF-alpha), interleukin (IL) 6 and IL15 relative to cyclophilin. Expression of leptin and adiponectin was not altered ($P > 0.05$) by exposure to 0.5% albumin. However TNF alpha mRNA abundance was elevated ($P < 0.001$) 1436 ± 134% within 1hr of exposure to albumin. Similarly, IL6 was elevated ($P < 0.001$) by greater than 1246 ± 286% within 1hr of incubation. In contrast, IL15 mRNA abundance was reduced ($P < 0.01$) by 62 ± 8% within 4hr. These data indicate that albumin alters the expression of specific adipocytokines. Secondly the data indicate that in vitro analysis of porcine adipocytokine expression may be confounded if albumin is utilized in the medium.

Key Words: adipose tissue, cytokine, swine

T133 Assisted reproductive technologies (ART) have a dramatic effect on cell proliferation in ovine fetal membranes (FM) during early pregnancy. P. P. Borowicz*[1], L. P. Reynolds[1], L. R. Coupe[1], G. Ptak[2], P. Loi[2], P. A. Scapolo[2], A. Cuomo[2], C. Palmieri[2], and A. T. Grazul-Bilska[1], [1]*North Dakota State University, Fargo*, [2]*Department of Comparative Biomedical Sciences, Faculty of Veterinary Medicine, University of Teramo, 64100 Teramo, Italy*.

Early pregnancy is a critical period for placentation and establishment of pregnancy. To determine the rates of cell proliferation in FM from pregnancies established after natural breeding (NAT) or after transfer of embryos created through ART, FM were collected on d 20 and 22 after breeding (n=4-6/day) from NAT pregnancies or after transfer of embryos created through natural breeding (ET), in vitro fertilization (IVF), or in vitro activation (IVA; parthenogenetic embryos). FM (n = 5-9/pregnancy type) were fixed in Carnoy's or formalin solution, embedded in paraffin and sectioned. Proliferating cells were identified using Ki67 immunohistochemstry, and nonproliferating cells were detected using nuclear fast red counterstaining. Micrographs of FM sections (n = 4-10/pregnancy type or day of pregnancy) were analyzed using image analysis (ImagePro Plus) to determine the labeling index (LI; proportion of proliferating cells out of the total cells per tissue area), which was similar for d 20 and 22 of NAT pregnancies. On d 20-22 of pregnancy, LI in FM was similar in NAT (21.5±3.9%) and ET (16.5±1.3%), which were greater ($P<0.01$) that IVF (9.9±1.7%) and IVA (8.5±1.4%) pregnancies. These data demonstrate that 1) the rate of cell proliferation is high in FM on d 20 and 22 of early pregnancy; 2) LI is similar for pregnancies from natural breeding or after ET from natural breeding on d 20 and 22; and 3) cellular proliferation in FM is decreased dramatically in embryos from ART such as IVF and IVA. Thus, application of ART may have profound effects on placentation and thus on pregnancy outcome. *Supported by USDA-NRI 2007-01215 to LPR and ATGB, and NIH P20 RR016741 (INBRE program of the National Center for Research Resource).*

Key Words: placenta, monoparental embryos, assisted reproduction

T134 SCD1 induction during early differentiation of bovine preadipocytes. L. Ma*, A. J. Lengi, and B. A. Corl, *Virginia Polytechnic Institute and State University, Blacksburg*.

Understanding the regulation of adipocytes and the role of stearoyl-CoA desaturase-1 (SCD1) in their differentiation is important for improving carcass quality. SCD1 catalyzes the synthesis of monounsaturated fatty acids. The first objective of this study was to determine the differentiation potential of primary bovine adipocytes in response to increasing passage number in culture. The second objective was to characterize SCD1 mRNA and protein induction during early differentiation of primary bovine adipocytes. Experiments were replicated at least 4 times using cells from different cattle. Preadipocytes were harvested from subcutaneous adipose tissue explants incubated in growth medium (DMEM with 10% FBS) for 12 d. After reaching confluence, differentiation medium was applied. To examine passage effects, cells were passed 1, 2, or 3 times before addition of differentiation media. Differentiation was assessed on d 8 using aP2 mRNA expression and glycerol-3 phosphate dehydrogenase (G3PDH) activity measured by microplate reader. With increasing pass number, mRNA abundance of aP2 decreased 50% (pass 2) and 90% (pass 3) compared to pass 1. G3PDH specific activity decreased about 30% (pass 2) and 70% (pass 3) compared to pass 1. To determine the induction of SCD1 expression during adipocyte differentiation, adipocytes were harvested 0, 1, 2, 3, and 8 d following induction of differentiation. mRNA expression of SCD1, aP2, and SREBP-1 were measured by real-time PCR, and SCD1 protein abundance was assayed by immunoblotting. The expression of aP2 mRNA increased 40-fold by d 1 and more than 400-fold by d 8 compared to d 0. SREBP-1 mRNA expression increased 4-fold on d 1 and was further increased almost 10-fold by d 8. For SCD1, mRNA abundance increased 4-fold at d 1 and was significantly increased more than 70-fold by d 8 compared to d 0. SCD1 protein increased more than 6-fold, 20-fold, and almost 900-fold by d 1, d 3, and d 8, respectively, compared to d 0. In conclusion, as passage number increases, the ability of preadipocytes to differentiate in culture decreases and during early differentiation of adipocytes, SCD1 is expressed and increases dramatically by d 8.

Key Words: adipocyte, differentiation

T135 Conjugated linoleic acid effects on adiposity are independent of spot 14 gene expression in mice. M. Hussein*, K. Harvatine, Y. Boisclair, and D. Bauman, *Cornell University*, *Ithaca, NY*.

Trans-10, cis-12 conjugated linoleic acid (CLA) decreases body fat accretion in several animal models when included as a dietary supplement. This CLA-mediated effect involves, but is not limited to, down-regulation of the expression and/or activity of transcription factors and key lipogenic enzymes regulating de novo lipid synthesis (DLS). Although its exact function is still unclear, thyroid hormone responsive spot 14 (S14) is expressed predominately in lipogenic tissues and is strongly correlated with DLS. Furthermore, S14 null mice exhibit impaired mammary DLS and CLA-induced reductions in synthesis of body fat (during growth) or milk fat (during lactation) correspond to a marked suppression of S14 expression. To test whether the effects on adiposity is mediated by S14, 9 wk old wild-type and S14-null mice (10/group) were orally dosed for 14 d with water (control) or 40 mg/d in two equal doses of CLA (50:50; t-10,c-12 and c-9,t-11 CLA isomers). Expression of selected transcription factors and lipogenic genes was assayed in white adipose tissue using quantitative RT-PCR. Epididymal and subcutaneous fat depots weighted slightly less for S14-null mice than wild type mice. In wild-type mice, the CLA supplement down regulated S14 expression in adipose tissue. CLA also reduced fat depot weights ($p<0.0001$), down regulated expression of fatty acid synthase, stearoyl-CoA desaturase, acetyl-CoA carboxylase, and sterol regulatory element binding protein-1c, and increased hepatic triglyceride content in wild- type mice; CLA had the exact same effects in S14-null mice. Taken together, the similarity of effects of CLA on adiposity and mRNA expression of selected lipogenic genes in S14-null and wild-type mice suggest that the effect of CLA on adipose tissue may be independent of the effects of CLA on S14 gene expression.

Key Words: CLA, Spot14, obesity

T136 The effect of KemTRACE® chromium propionate supplementation on global gene expression in adipocytes of finishing pigs. L. Wonderling*[1], J. Hahn[1], M. Spurlock[2], and A. Jourdan[1], [1]*Kemin Industries*, *Des Moines, IA*, [2]*Iowa State University*, *Ames*.

Supplementation with KemTRACE® Chromium Propionate (CrProp) has been demonstrated to benefit swine growth and health, but the molecular mechanism for these effects are largely unknown. To better understand KemTRACE® CrProp's molecular role, a global gene expression study was performed. KemTRACE® CrProp supplements were fed to 8 pigs over a 2 month period corresponding to the finishing stage of growth. A second set of 8 pigs were used as non-supplemented control animals. Adipose tissue was removed from the shoulders of all pigs prior to supplementation, and following 1 and 2 months of supplementation. RNA was isolated from the tissue samples and used in hybridization studies with the Affymetrix porcine microarray. Lists of differentially expressed genes for the control and treated groups were compiled ($\geq$ 1.5-fold, $p \leq 0.001$). The control gene list was subtracted from the treated group gene list to identify genes whose expression responded to KemTRACE® CrProp treatment over time. When comparing gene expression at the different study time points, CrProp affected the expression of 133 genes in the 1st month, 527 genes in the 2nd month, and 1328 genes if the pre-treatment to post-treatment gene expression is compared. The overlap of these gene lists was examined and the results suggest that 84 genes were specific to the 1st month of treatment, 232 genes were specific to the 2nd month of treatment, and 997 genes were only affected by CrProp when comparing the pre-treatment to the end of supplementation. Gene clustering was performed using NIH DAVID. Gene clusters affected by CrProp included those involved in central immune pathways, cell proliferation/apoptotic pathways, and in the development of muscle, extracellular matrix, bone and organs. Interestingly, the expression of several genes involved in key steps of insulin signaling was affected by CrProp supplementation including the PI3K, GLUT4, and AMPK genes. These results may indicate suppressed development of adipocytes in pigs supplemented with KemTRACE® CrProp.

Key Words: chromium, nutrition, adipocytes

T137 Expression of microRNA in bovine preadipocytes and adipocytes. S. L. Pratt, A. P. Burns, and S. K. Duckett*, *Clemson University*, *Clemson, SC*.

It has recently been demonstrated that adipogenesis involves microRNA (miRNA). miRNA are small non-coding inhibitory RNA that are present and expressed in the cells of all plants and animals examined to date, and regulate gene expression post-transcriptionally by either translational repression or RNA interference. To date, little information is available concerning either the identification or function of miRNA in bovine adipocyte differentiation. The objective of this study was to evaluate the expression profile of miRNA in pre- and differentiated adipocytes utilizing miRNA microarrays. Bovine preadipocyte cell lines 7-01 and 7-02, generated from subcutaneous adipose tissue obtained from 18 mo old steers were cultured to confluence and held 2 d in Dulbecco's modified eagles medium (DMEM) containing 10% fetal calf serum (FCS), and 2X antibiotiotic/antimycotic (AB/AM). Cells were differentiated using a two step process by culture for 2 d in DMEM containing 5% FCS, 2X AB/AM, Insulin at 2.5 µ/ml, 0.25 µM dexamethasone, 5 µM troglitasone and 0.5 µM isobutylmethylxanthine for followed by 4 d incubation in DMEM containing 5% FCS, 2X AB/AM, Insulin, and troglitasone. tcRNA was isolated for each cell line at 2 d confluence (Control), and after 2 d (D2) and 6 d (D6) of differentiation, and were then used in a 3 x 3 microarray using control, D2 and D6 samples. Data were subjected to t-test analysis and six miRNA were determined to be differentially regulated. For preadipocytes and adipocytes, 167 and 182 total miRNA were detected, respectively. The miRNA let-7a, let-7b, and let-7c were upregulated ($P<0.01$), while the miRNA, miR-21, miR-221, and the miR-222 were down regulated ($P<0.01$). All miRNA that were differentially expressed have been detected by RT-PCR except for let-7b. The let-7 miRNA have roles in differentiation of multiple cell and tissue types, while the miR-21, miR-221, and miR-222 all have been implicated in cell cycle regulation.

Key Words: microRNA, adipocyte, differentiation

T138 Characterization of ovine fetal heart gene expression during fetal growth restriction. K. A. Partyka*[1], J. S. Barry[2], R. V. Anthony[1,2], and H. Han[1], [1]*Colorado State University*, *Fort Collins*, [2]*University of Colorado Health Sciences Center*, *Aurora*.

Growth restricted fetuses (FGR) are hypoglycemic, hypoxemic and show increased systemic vascular resistance, all of which could impact heart development. Objective: To examine cardiac gene expression at two gestational ages in ovine FGR fetuses. Single bearing ewes were placed in a hyperthermic (HT) environment at 35 days gestation (dGA) (40°C 12 hrs/day and 35°C 12 hrs/day; n=11) to induce FGR. The ewes were maintained in the HT environment until studied at 90 dGA (n=6), or 120 dGA (n=5) then placed in control conditions until studied at 135 dGA. Pair-fed control (TN; 20°C ± 2°C 24 hrs/day; n=14) ewes

were also studied at 90 dGA (n=5) or 135 dGA (n=9). Fetal and heart weights were measured; the heart was sectioned into right ventricle (RV) and left ventricle plus septum (LV+S), snap frozen and stored at -80°C until analysis. Quantitative real-time PCR was used to determine mRNA concentrations for endothelial nitric oxide synthase (eNOS), type 1 (AT1) and 2 (AT2) angiotensin II receptor, angiopoietin 1 (Ang1) and 2 (Ang2), tunica interna endothelial cell kinase 2 (Tie2), vascular endothelial growth factor (VEGF), VEGF receptor 1 (VEGFR1) and 2 (VEGFR2), glucose transporter 1 (GLUT1) and 4 (GLUT4), insulin receptor β (IRβ), myosin heavy chain α (MHCα) and β (MHCβ). Data was analyzed by Students t-test. Neither fetal or heart weight was impacted by HT at 90 dGA, whereas both fetal (1529 g vs 3294 g; P≤0.01) and heart weight (13.6 g vs 29.1 g; P≤0.01) were reduced by HT at 135 dGA. HT 90 dGA showed an increased RV AT2 (P≤0.05) and AT2/AT1 ratio (P≤0.05), LV+S MHCβ (P≤0.05) and MHCβ/MHCα ratio (P≤0.01). HT 135 dGA, LV+S VEGF (P≤0.05) and RV VEGFR1 (P≤0.05) mRNA concentrations were greater. The fetal myocardium adapts during FGR by altering gene expression that may represent delayed maturation at 90 dGA and signal for increased myocardial angiogenesis near term, both changes that could be detrimental later in life. *Supported by NIH grant HD043089.*

Key Words: heart, sheep, fetal growth restriction

T139 Development of a protocol for staining BrdU-labeled cells within cryosections of bovine mammary tissue that is suitable for subsequent transcriptome analysis. R. K. Choudhary*[1], K. M. Daniels[2], C. Clover[2], and A. V. Capuco[2,1], [1]*University of Maryland, College Park*, [2]*Bovine Functional Genomics Laboratory, USDA-ARS, Beltsville, MD*.

Bromodeoxyuridine (BrdU) is a thymidine analog that is incorporated into the DNA of proliferating cells. Immunostaining of BrdU-labeled cells within a histological section requires heat or chemical-mediated antigen retrieval to open the dsDNA and expose the BrdU antigen. We found that in cryosections such treatments, while they do permit staining of the tissue, preclude its use in further gene expression experiments. With cryosections, long antibody incubations and several washing steps, lead to extensive RNA degradation and undesired RNA elution into wash buffers. We have developed a protocol for immunolocalization of BrdU-labeled cells in unfixed cryosections of bovine mammary tissue that does not require harsh DNA denaturation and minimizes RNA degradation and elution into wash buffers. This protocol uses an initial acetone treatment (2 min) followed staining with methyl green (0.5%, 2 min) to stabilize macromolecules, antigen retrieval with deionized formamide (70% in nuclease-free phosphate buffered saline, 5 min incubation), short (10 min) antibody incubations in the presence of RNase inhibitors, and minimal washing, to facilitate recovery of RNA from stained sections. Total RNA was obtained by column extraction (RNeasy Micro kit; Qiagen, Valencia, CA) of cell lysates from sections. Utility of the isolated RNA for gene expression analysis was confirmed using gene-specific primers and quantitative RT-PCR. Furthermore, RNA quality was evaluated by micro-fluidic electrophoresis (Agilent BioAnalyzer) and acceptable RNA integrity numbers (RIN 6.0) were obtained. Staining intensity obtained with this BrdU labeling protocol was similar to that obtained using conventional immunohistochemistry protocols. When coupled with laser microdissection and RNA amplification, this immunostaining protocol should provide a means for evaluating genes expressed by BrdU-labeled cells that are extracted from a complex tissue section.

Key Words: BrdU staining, gene expression, laser microdissection

T140 Analysis of protein oxidation in serum of fetal and newborn piglets and the influence of iron dextran on induction of protein carbonyls. T. J. Caperna*, A. E. Shannon, T. G. Ramsay, L. A. Blomberg, and W. M. Garrett, *USDA/ARS, Beltsville, MD*.

Oxidation of serum proteins leads to non-reversible carbonyl formation which alters their function and has long been associated with stress-related disease processes. The objective of this study was to identify serum protein biomarkers of metabolic stress in baby pigs. Protein carbonyls in serum, were converted to dinitrophenyl (DNP) derivatives with DNP-hydrazine, precipitated with TCA, extracted in ethyl acetate:ethanol (1:1) and quantified by spectrophotometry. DNP-labeled proteins were then separated by 2D PAGE, electro-blotted onto PVDF membranes and visualized by western immunoblot using polyclonal DNP antiserum. Proteins which reacted positively in western blots were extracted from duplicate 2D gels, digested with trypsin and identified by MALDI-TOF mass spectrometry (MS). At birth, significant amounts of oxidized proteins were readily determined in piglet serum (~1 nmole/mg protein). Fetuses at 50 and 110 d of gestation were also evaluated and found to have potentially higher levels of protein carbonyls, than newborns. We then determined that the standard iron dextran treatment used to prevent anemia in pigs (100 mg iron given intramuscularly), was associated with a two-fold increase (P<.05) in oxidized proteins by 48 hrs, if given on postnatal day 1 (n = 7 pigs/group). Oxidized proteins identified by MS included; albumin, transferrin, alpha fetoprotein, haptoglobin, immunoglobulins and fetuin. Postponement of iron treatment until day 3 also resulted in an increase in protein carbonyls, but at attenuated levels (P<.05) compared to piglets treated on day 1. Therefore, adjusting the timing of treatment should reduce the impact of iron dextran as a source of oxidative stress in newborn pigs. These results indicate that the sensitivity and simplicity of the analysis to evaluate protein carbonyls, which was routinely performed using one mg of serum protein, may be a relatively non-invasive, rapid and useful tool to monitor oxidative stress associated with neonatal mortality in the pig.

Key Words: protein carbonyl, oxidative stress, mass spectrometry

T141 AMP-activated protein kinase γ3 subunit mutation in transgenic mice corresponding to RN- allele in pigs inhibits adipogenesis. J. X. Zhao*, X. Yan, J. F. Tong, M. J. Zhu, and M. Du, *University of Wyoming, Laramie*.

Due to genetic selection for lean growth, Rendement Napole (RN-) allele has increased in frequency in Hampshire pigs. Recently, the RN- allele was identified as a R225Q mutation in AMP-activated protein kinase (AMPK) γ3 subunit. The mechanisms linking AMPK and leanness in RN- pigs remain largely undefined. Since AMPK is a kinase controlling energy metabolism, we hypothesized that AMPK mutation in RN- pigs inhibits adipogenesis, especially when fed a high-energy diet, and diverts this excessive energy for lean growth. The objective of this study was to study the role of AMPK in adipogenesis and myogenesis of RN- pig using a transgenic mouse model. Wild-type (WT) and transgenic mice carrying the R225Q mutation (RN) were assigned to four groups according to the genotype and diet: control group with normal energy diet (WTnormal, n=6), control group with high energy diet (WTfat, n=6), mutated group with normal energy diet (RNnormal, n=6) and mutated group with high energy diet (RNfat, n=6). One month-old mice were fed the treatment diets for one month. Then, mice were euthanized, and carcass, muscle samples, gonad and skin fat were collected and weighed. In WTnormal, the gonad fat weight was 0.91±0.14 g and skin fat weight was 0.68±0.08 g, versus 0.80±0.10 g and 0.58±0.05 g in RNnormal, without significant difference. However, difference was

observed between WTfat and RNfat mice. In WTfat, the gonad fat and skin fat weights were 1.16±0.13 g and 0.74±0.07 g versus 0.77±0.08 g and 0.56±0.04 g respectively ($P < 0.05$) for RNfat mice. Intriguingly, RN mutation enhanced feed consumption ($P < 0.05$) of mice both in normal- and high-energy diets. The high feed intake but increased leanness indicates that RN- allele enhances energy expenditure which warrants further studies.

Key Words: AMPK, skeletal muscle, adipogenesis

T142 Growth hormone does not stimulate IGF-I mRNA expression in bovine skeletal muscle, myoblasts, or myotubes. X. Ge and H. Jiang*, *Virginia Polytechnic Institute and State University*, *Blacksburg*.

Growth hormone (GH) is a major regulator of muscle growth in animals, including cattle. The growth-stimulating effect of GH had long been thought to be mediated by circulating IGF-I produced from the liver. However, recent knockout mouse studies support the concept that IGF-I produced in the skeletal muscle and IGF-I-independent mechanisms mediate the effect of GH on growth. To clarify the mechanism by which GH stimulates skeletal muscle growth in cattle, we determined whether GH increased IGF-I mRNA expression in bovine skeletal muscle in vivo and bovine myoblasts and myotubes in vitro. Five nonlactating, nonpregnant beef cows were administrated subcutaneously with 500 mg of recombinant bovine GH formulated for slow release, and longissimus muscle biopsies and blood samples were taken seven days before and seven days after GH administration. Serum IGF-I concentrations, measured by an ELISA, were four times higher after GH injection than before GH injection ($P < 0.01$). Skeletal muscle IGF-I mRNA abundance, measured by real-time RT-PCR, was not different between before and after GH injection ($P > 0.1$). Western blotting analyses showed that GH administration increased the levels of phosphorylated JAK2 and STAT5, known protein components of GH signaling, in the skeletal muscle ($P < 0.05$). In the in vitro experiments, myoblasts were isolated from bovine skeletal muscle and were allowed to proliferate or induced to form myotubes in culture. Growth hormone at the concentrations of 10 ng/ml and 100 ng/ml did not affect IGF-I mRNA expression in either the myoblasts or myotubes ($P > 0.1$). These in vitro data echoed the earlier in vivo observation that GH did not increase IGF-I mRNA expression in the skeletal muscle. Taken together, the data of this study demonstrate that GH does not increase skeletal muscle IGF-I mRNA expression in cattle and suggest that GH stimulation of muscle growth in cattle is not mediated by the locally produced IGF-I.

Key Words: growth hormone, skeletal muscle, IGF-I

T143 Early-weaning down-regulates the expression of aminopeptidase N gene in the jejunum of the piglet. D. Lackeyram*, T. Archbold, K. C. Swanson, and M. Z. Fan, *University of Guelph*, *Guelph, Ontario, Canada*.

Aminopeptidase N (APN) cleaves neutral AA from oligopeptides during digestion in the small intestine. The objectives of this study were to examine the responses of APN activity and protein abundances in the partitioned mucosal homogenate (H), intracellular soluble and the apical membrane (M) fractions as well as APN mRNA abundance during early-weaning in comparison with suckling pigs. A total of 20 Yorkshire piglets, 10 suckling (SU) and 10 early-weaning (WN) with an average BW of 3 kg at the age of 10 d, were used in this study. Weanling piglets were fed a corn and SBM-based diet for 12 d. Proximal jejunal samples from both SU and WN groups were collected. L-alanine-P-nitroanilide hydrochloride (0-16 mM) was used in the enzymatic kinetic experiments. Abundances of APN protein and mRNA were analyzed by Western blot and the real time RT-PCR using β-actin as the housekeeping gene. The jejunal APN maximal specific activity (μmol/mg protein.min) was decreased ($P<0.05$) in weaning piglets (H: WN, 0.122±0.01 vs. SU, 0.172±0.01; and M: WN, 0.5479±0.02 vs. SU, 0.721±0.01). There were decreases ($P<0.05$) in the APN protein abundance (arbitrary units) for the WN group in the jejunal fractions (H: WN, 0.655±0.03 vs. SU, 0.992±0.05; and M: WN, 0.232±0.02 vs. SU, 0.515±0.01). There were correlations ($P<0.05$) between the APN maximal specific activity and its protein abundance (H, $r = 0.87$; M, $r = 0.91$, $n=20$). Furthermore, early-weaning decreased ($P<0.05$) the relative abundance of APN mRNA by 39% (WN, 0.304±0.02 vs. SU, 0.498±0.02). The APN mRNA abundance was also correlated ($P<0.05$, $n=20$) with its protein abundances in both homogenate (H, $r = 0.82$) and membrane (M, $r = 0.86$) fractions. We conclude that the reduced activity of APN during early-weaning can be attributed to a down-regulation of the APN gene at the transcriptional and translational levels.

Key Words: aminopeptidase N, gene expression, weanling pigs

Horse Species

T144 Influence of extension on the stock-type western pleasure jog. M. Nicodemus* and J. Williams, *Mississippi State University*, *Mississippi State*.

One of the most popular performance classes for the stock-type breed is the western pleasure class. Previous research of the stock-type western pleasure gaits found the jog and lope were performed as 4-beat stepping gaits with periods of quadrupedal support. To encourage breeds to perform with more forward motion, stock-type breed associations added to the class requirements extension of the jog. Study objectives were to determine the influence of extension on the temporal variables of the stock-type jog. 5 registered stock-type horses showing in a western pleasure class were filmed at 60 Hz. Strides of both the jog and extended jog that were judged as desirable by carded judges were evaluated using frame-by-frame analysis. Means (SD) were determined for 5 strides for each horse for both gaits with variables given as % of stride. Effect of extension for each variable was tested using a one-way analysis of variance ($p<0.05$). Velocity (Jog: 1.5±0.1 m/s, Extended: 2.2±0.2 m/s) and stride length (Jog: 1.6±0.1 m, Extended: 2.5±0.2 m) and duration (Jog: 917±67 ms, Extended: 873±42 ms) were not significantly influenced by extension ($p>0.05$). Majority of stride for both the jog (Right: Fore=67±2%, Hind=63±6%; Left: Fore=66±3%, Hind=64±4%) and extended jog (Right: Fore=66±4%, Hind=60±3%; Left: Fore=64±2%, Hind=61±3%) was spent in stance with hind stance shortening with extension ($p<0.05$). Rhythm changed from 4 to 2 beats as extension created diagonal limb pairing during the extended jog (Jog: Right Hind-Left Fore Advanced Placement=4±1%, Left Hind-Right fore=4±1%; Extended: Right Hind-Left Fore Advanced Placement=0±0%, Left Hind-Right Fore Advanced Placement=0±0%). While both gaits were performed as stepping gaits with the greatest % of stride spent in diagonal bipedal support (Jog: 63±7%, Extended: 70±5%), extension shortened limb support during tripedal support with 2 hind limbs (Jog: 5±3%,